PATENT

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

Applicant: P. A. Billing-Medel, *et al.*

Serial No.: 09/049,695

Filed:

For: REAGENTS AND METHODS USEFUL FOR DETECTING DISEASES OF THE GASTROINTESTINAL TRACT

Examiner: K. Canella

Group Art Unit: 1642

Attorney Docket No.: 6066.US.P1

Date: July 23, 2001

Certificate of Mailing under 37 CFR §1.8(a): I hereby certify that this paper (al ng with any paper referred to as being attached or enclosed) is being deposited with the United States Postal Service with sufficient postage as First Class Mail addressed to:

Box CPA
Assistant Commissioner for Patents
Washington, D.C. 20231

Date of Deposit: July 23, 2001

Ruth Pe Palileo

DECLARATION OF PHILIP HEMKEN UNDER 37 C.F.R 1.132

Box CPA
Assistant Commissioner of Patents
Washington, D.C. 20231

Dear Sir:

I, the undersigned, declare as follows:

1. I am one skilled in the art of cancer diagnostics. I have a Ph.D. in Molecular, Cellular and Developmental Biology from Iowa State University. I have an M.A. in Biotechnology from Washington University in St. Louis. I further have a B.S. in Microbiology from Iowa State University.

2. I was a Postdoctoral Fellow in the Laboratory of Dr. Andrei Mirzabekov at Argonne National Laboratory.

3. I have four years of research and development experience in the cancer diagnostic industry. Much of my work has involved the discovery and validation of novel cancer markers to improve the accuracy of diagnosing the onset of cancer. (*See* Attachment A, which is my Curriculum Vitae).

4. I have read and am familiar with the Patent Office Action dated November 22, 2000 in the parent application serial no. 09/049,065.

5. I have read and am familiar with the Advisory Action dated May 21, 2001 in the parent application and with the utility rejection under 35 U.S.C. 101 applied against the parent application.

6. I have reviewed the attached data, in the form of FIG. 1, relating to the gene product designated in the instant application as CS197.

7. In order to provide the attached data, reverse transcriptase-polymerase chain reaction (RT-PCR) was performed to evaluate the expression of CS197 in two normal colon tissue samples and two tumor colon tissue samples. Total RNA was isolated from the four tissue samples and cDNA was prepared with 1 μg total RNA using random hexamer primers and the First Strand cDNA Synthesis Kit (from Invitrogen). cDNA (1 μl) was used for PCR of the colon tissues. The primers are, from the 5' end, forward 87 and reverse 188. This results in a 100 bp product. The amplification conditions comprised of 40 cycles of 95°C for 15 sec and 60°C for 45 sec. The PCR products were separated by 4% agarose gel electrophoresis and the CS197 bands were scanned from left to right.

8. FIG. 1 reports the area of individual peaks and the average of 2 peaks (i.e., the average of the two CT1 peaks, the two CT2 peaks, etc).

9. As seen in FIG. 1, scans of "TUMOR" samples 1, 2, 3 and 4 are considerably higher than the scans of "NORMAL" samples 5, 6, 7 and 8.

10. Thus, CS197 expression is higher in tumor colon tissue (samples 1-4) than in normal colon tissue (samples 5-8).

11. On average, the expression of CS197 in tumor colon tissue is 2.0 to 2.8 times higher than its expression in normal colon tissue.

12. Thus, CS197 expression is upregulated in tumor colon tissue samples versus normal colon tissue samples. Such upregulation indicates that a disease has altered the polynucleotides so that their expression in their normal host tissue is disrupted in tumor tissue.

13. Thus, CS197 is able to distinguish between normal colon tissue and tumor colon tissue.

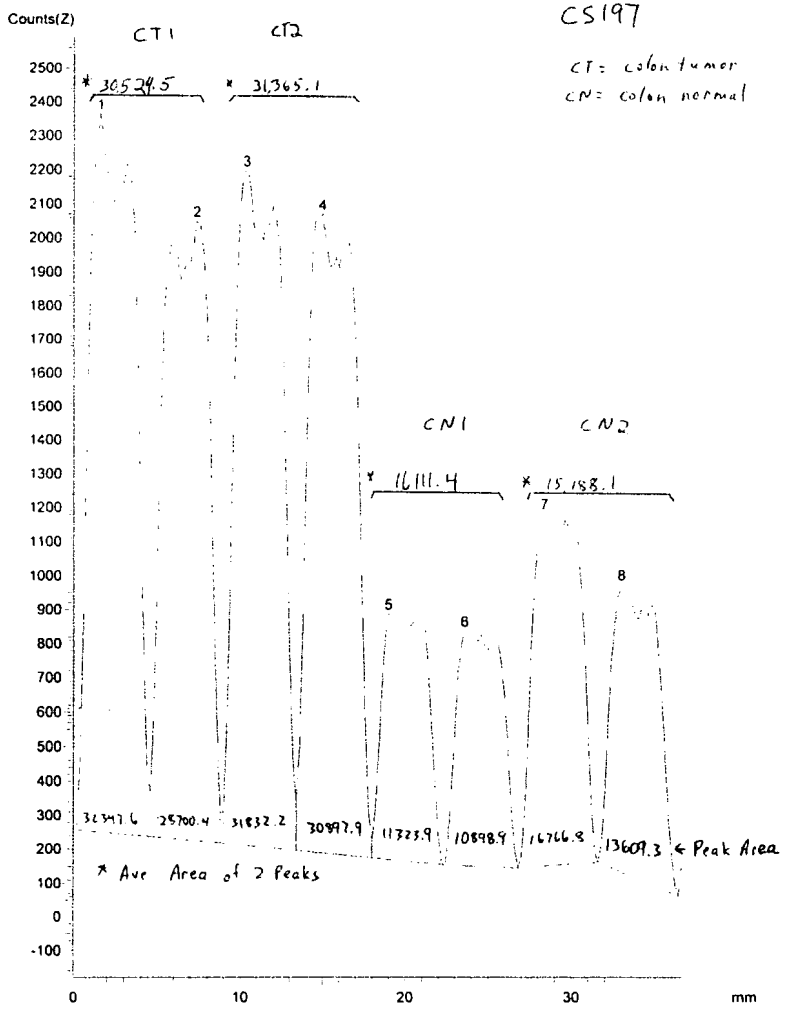
14. Clearly CS197 is characteristic of a tissue-specific marker and able to act as a cancer diagnostic as evidenced by the above data.

15. I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that the statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both, under § 1001 of Title 18 of the United States Code and such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Philip Hemken, Ph.D.

7/27/01
Date

FIG. 1



Peaks	Area of Individual Peaks		*Average area of 2 peaks	
1	CT1	32347.6	CT1	30524.0
2	CT1	28700.4		
3	CT2	31832.2	CT2	31365.1
4	CT2	30897.9		
5	CN1	11323.9	CN1	11111.4
6	CN1	10898.9		
7	CN2	16766.8	CN2	15188.1
8	CN2	13609.3		